New publications in the PARATUBERCULOSIS database (1211-1213)

Mycobacterium avium subsp hominissuis Infection in a Captive-Bred Kiang (Equus kiang)
Journal of Comparative Pathology, 146, 372-377

Equids are considered highly resistant to mycobacterial infections and clinical cases have been described in domestic horses only. Mycobacterium bovis is the most common species reported, although a single report exists of disease due to definitively diagnosed infection with Mycobacterium avium subsp. hominissuis in two domestic horses. This is the first report of a mycobacterial infection in a kiang (Equus kiang), or indeed any wild equid. The animal had chronic loss of condition and serum biochemical changes suggestive of liver disease and chronic infection. Further investigation showed a chronic granulomatous enteritis, lymphadenitis and hepatitis with focal granulomatous pneumonia due to systemic infection with M. avium subsp. hominissuis. The distribution and severity of the lesions suggested that the route of infection was alimentary. (C) 2011 Elsevier Ltd. All rights reserved

Production and proteomic characterisation of purified protein derivative from Mycobacterium avium subsp paratuberculosis
Proteome Science, 10, Background: Effective diagnosis of Johne's disease (JD), particularly at the stage of early subclinical infection, remains one of the greatest challenges for the control of JD worldwide. The IFN-gamma test of cell mediated immunity is currently one of the most suitable diagnostics for subclinical infections, however a major limitation of this test is the lack of a standardised purified protein derivative (PPD) antigen (also referred to as Johnin PPD or PPDj). While attempting to replace PPDj with more specific individual antigens is an attractive proposition, bacterial culture derived PPDj remains the most effective antigen preparation for the diagnosis of subclinical JD. It may be possible to increase the reproducibility and specificity of PPDj preparations by further characterising and standardising the PPDj production. Results: Using a standardised protocol, five in-house preparations of PPDj were prepared from cultures of Mycobacterium avium subsp. paratuberculosis (MAP). Compared to PPDs obtained from other institutes/laboratories, these preparations appeared to perform similarly well in the IFN-gamma test. Although the broad proteomic composition of all PPDj preparations was remarkably similar, the absolute abundance of individual proteins varied markedly between preparations. All PPDj preparations contained common immunogenic proteins which were also observed in PPD preparations from Mycobacterium avium subsp. avium (PPDa) and Mycobacterium bovis (PPDb). Temporal difference in protein secretion of in vitro cultured MAP was observed between 20 and 34 weeks suggesting that the age of MAP culture used for PPDj preparations may markedly influence PPDj composition. Conclusions: This study describes a protocol for the production of PPDj and its subsequent proteomic characterisation. The broad proteomic composition of different preparations of PPDj was, for the most part, highly similar. Compositional differences between PPDj preparations were found to be a direct reflection of genetic differences between the MAP strain types used to produce these preparations and the age of MAP cultures they were derived from. A number of conserved immunogenic proteins, such as members of the cutinase-like protein family, were found to be more abundant in PPDj compared to PPDa and should be considered as possible diagnostic antigens for the future
Aims: To compare three decontamination methods applied to paucibacillary samples for primary isolation of Mycobacterium bovis from suspect lesions. Tuberculosis caused by Myco. bovis is an important infectious disease of cattle in Brazil and also has zoonotic potential. Although a national campaign based on testing and slaughtering cattle has achieved good results, there is a strong need to develop better diagnostic methods to identify cattle with recent infections harbouring few bacilli.

Methods and Results: A dairy herd (274 adult crossbred cows) located in the state of Rio de Janeiro was tested for tuberculosis with both single intradermal tuberculin test and comparative intradermal tuberculin Lest. Reactive cows (n = 27, 9.8%) were slaughtered and suspect lesions were collected (one sample per cow). Samples considered paucibacillary (based on microscopy) were decontaminated with 0.75% hexadecylpyridinium chloride (HPC), 400 sodium hydroxide (Petroff) or 600 sulphuric acid. Using these methods, 10, five and six, respectively, of the 27 samples yielded positive cultures. Overall, Myco. bovis was isolated from 14 of 24 cows. Although the HPC method resulted in isolation of more Myco. bovis strains than either Petroff or sulphuric acid methods (P = 0.015), it did not result in the recovery of Myco. baits from all samples. However, using both HPC and 6% sulphuric acid methods for decontamination was possible to identify 13 of 14 (92.9%) of infected cows. Conclusions: At least two methods should be used concurrently for primary isolation of Myco. bovis from bovine tissues, particularly for paucibacillary samples. Significance and Impact of the Study: Detection of low numbers of Myco bovis in tissue is an important goal in optimizing the detection of bovine tuberculosis and should assist in identification of infected cattle, in particular, those with few Myco, bovis bacilli. This was apparently the first study comparing three decontamination methods for the detection of Myco bovis in paucibacillary samples from naturally infected cattle.
Crohn's disease carrying the Leu1007 frameshift mutation of NOD2, we showed that (i) both NOD2 dependent and independent signalling (appearing TLR2 mediated) occurred for PGN upregulation of PD-L1 (ii) upregulation is lost in response to MDP in patients with the homozygous mutation and (iii) PD-L1 upregulation was unaffected in patients with heterozygous mutations as previously reported for cytokine responses to MDP. The uptake of PGN and its cleavage products by the intestinal mucosa is well recognised and further work should consider PD-L1 upregulation as one potential mechanism of the commensal flora-driven intestinal immuno-tolerance. Indeed, recent work has shown that loss of PD-L1 signalling in the gut breaks CD8(+) T cell tolerance to self antigen and leads to severe autoimmune enteritis. (C) 2012 Elsevier Inc. All rights reserved

**Altered Oligosaccharide Structures Reduce Colitis Induction in Mice Defective in beta-1,4-Galactosyltransferase**

Gastroenterology, 142, 1172-1182

BACKGROUND & AIMS: Oligosaccharide modifications induce various functional changes in immune cells. The galactose-deficient fraction of fucosylated IgG oligosaccharides is increased, whereas that of beta-1,4-galactosyltransferase I (B4GalTI) is reduced, in patients with Crohn's disease. We investigated the role of oligosaccharide modification in the pathophysiology of colitis using B4galt1-deficient mice. METHODS: Colitis severity was compared between B4galt1(+/+) and B4galt1(+/-) mice. B cells isolated from B4galt1(+/+) and B4galt1(+/-) mice were adoptively transferred to recombination activating gene 2 (-/-) mice, in which colitis was induced by administration of CD4(+)CD62L(+) T cells. Cell-surface glycan profiles were determined by lectin microarray analysis. Cytokine production was determined in a coculture of various types of cells isolated from either B4galt1(+/+) or B4galt1(+/-) mice. RESULTS: Colitis induction by dextran sodium sulfate or trinitrobenzene sulfonic acid was significantly reduced in B4galt1(+/-) mice, which had galactose deficiency in IgG oligosaccharides (similar to patients with Crohn's disease) compared with B4galt1(+/+) mice. Amelioration of colitis was associated with increased production of interleukin-10 by macrophages in B4galt1(+/-) mice. Colitis induction in recombination activating gene 2(-/-) mice by administration of CD4(+)CD62L(+) T cells was reduced by cotransfer of B4galt1(+/+) cells, but not from B4galt1(+/-) mice. Lectin microarray analysis revealed increased expression of polylactosamines on B4galt1(+/-) cells and macrophages, compared with B4galt1(+/-) cells. The production of interleukin-10 from macrophages was induced via their direct interaction with B4galt1(+/-) B cells. CONCLUSIONS: Altered oligosaccharide structures on immune cells modulate mucosal inflammation. Oligosaccharides in immune cells might be a therapeutic target for inflammatory bowel diseases.

**Crohn's disease-associated polymorphism within the PTPN2 gene affects muramyl-dipeptide-induced cytokine secretion and autophagy**

Inflammatory Bowel Diseases, 18, 900-912

Background: The single nucleotide polymorphism (SNP) rs2542151 within the gene locus encoding protein tyrosine phosphatase non-receptor type 2 (PTPN2) has been associated with Crohn's disease (CD), ulcerative colitis (UC), type-I diabetes, and rheumatoid arthritis. We have previously shown that PTPN2 regulates mitogen-activated protein kinase (MAPK) signaling and cytokine secretion in human THP-1 monocytes and intestinal epithelial cells (IEC). Here, we studied whether intronic PTPN2 SNP rs1893217 regulates immune responses to the nucleotide-oligomerization domain 2 (NOD2) ligand, muramyl-dipeptide (MDP). Materials and Methods: Genomic DNA samples from 343 CD and 663 non-IBD control patients (male and female) from a combined German, Swiss, and Polish cohort were genotyped for the presence of the PTPN2 SNPs, rs2542151, and rs1893217. PTPN2-variant...
rs1893217 was introduced into T84 IEC or THP-1 cells using a lentiviral vector. Results: We identified a novel association between the genetic variant, rs1893217, located in intron 7 of the PTPN2 gene and CD. Human THP-1 monocytes carrying this variant revealed increased MAPK activation as well as elevated mRNA expression of T-bet transcription factor and secretion of interferon-? in response to the bacterial wall component, MDP. In contrast, secretion of interleukin-8 and tumor necrosis factor were reduced. In both, T84 IEC and THP-1 monocytes, autophagosome formation was impaired. Conclusions: We identified a novel CD-associated PTPN2 variant that modulates innate immune responses to bacterial antigens. These findings not only provide key insights into the effects of a functional mutation on a clinically relevant gene, but also reveal how such a mutation could contribute to the onset of disease. (Inflamm Bowel Dis 2011;